

**Structural and functional analysis of the *Bacillus anthracis* nutrient
germinant receptor proteins**

by

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DEDICATION

To my grandmothers, Mary Koudelka and Faye Jeanell Wilson.

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TABLE OF CONTENTS

DEDICATION	ii
ACKNOWLEDGEMENTS	iii
LIST OF FIGURES	vii
ABSTRACT	ix
CHAPTER 1: INTRODUCTION	1
1.1. Spore-forming Pathogens	1
1.2. Anthrax	2
1.2.1. Routes of Infection	4
1.2.2. Mechanisms of Virulence	5
1.3. Spore Structure and Formation	7
1.3.1. Transcriptional Regulation of Sporulation	8
1.3.2. Spore Core	11
1.3.3. Cortex	14
1.3.4. Coat and Exosporium	15
1.4 Germination	17
1.5. Nutrient Germinant Receptors	21
1.5.1. nGR genes	21
1.5.2. Receptor Hypothesis	24
1.5.3. Complex Formation	25
1.5.4. Signal Transduction	26
1.5.5. Bacillus anthracis nGR germinant responses	28
1.5.6. inter-nGR Interactions	28
1.6. Dissertation Project	30
CHAPTER 2: MEMBRANE TOPOLOGY OF THE BACILLUS ANTHRACIS GERH GERMINANT RECEPTOR PROTEINS	31
Abstract	31

2.1. Introduction	33
2.2 Materials and Methods	36
2.3. Results	44
2.3.1 Bacillus anthracis GerH overexpression in vegetative bacilli	44
2.3.2. GerH _C topology	45
2.3.3. GerH _B topology	49
2.3.4. GerH _A topology	56
2.4. Discussion.....	63
CHAPTER 3: FUNCTIONAL INTERACTIONS BETWEEN THE BACILLUS ANTHRACIS NUTRIENT GERMINANT RECEPTOR PROTEINS	71
Abstract.....	71
3.1. Introduction	73
3.2. Materials and Methods	75
3.3. Results	80
3.3.1. All three nGR subunits are required for germination.....	80
3.3.2. Complementation of nGR subunit deletions	80
3.3.3. The A-type subunits are partially interchangeable.....	92
3.3.4. The A-type subunits play a role in determining germinant specificity.....	94
3.3.5. Inter-nGR Interactions.....	95
3.4. Discussion.....	97
CHAPTER 4: DISCUSSION AND FUTURE DIRECTIONS.....	103
4.1. Summary.....	103
4.2. nGR Complex Formations	104
4.3. Role of the nGR subunits in triggering germination	106
4.3.1. B-type subunits	106
4.3.2. A-type subunits	109

4.3.3. C-type subunits	110
4.3.4. Functional redundancy of the nGR subunits	112
4.4. Inter-nGR interactions.....	113
4.5. Implications	114
REFERENCES	116

LIST OF FIGURES

Figure 1.1.	Stages of <i>B. anthracis</i> sporulation.....	10
Figure 1.2.	Spore Structure	13
Figure 1.3.	Model of <i>B. anthracis</i> germination.....	20
Figure 2.1.	Discrimination between intracellular and extracellular protein domains in <i>B. anthracis</i>	46
Figure 2.2.	Analysis of GerH _B topology	53
Figure 2.3.	Representative samples of GerH _B fusion protein analysis	54
Figure 2.4.	Analysis of GerH _A topology.	60
Figure 2.5.	Representative samples of GerH _A fusion protein analysis	61
Figure 2.6.	Topology Models of the <i>B. anthracis</i> GerH _A and GerH _B	64
Figure 2.7.	Current model of the nutrient germinant receptors in the inner membrane of the spore	66
Figure 2.8.	Alignment of <i>B. anthracis</i> GerH _A and <i>B. subtilis</i> GerA _A amino acid sequences.	69
Figure 3.1.	All three nGR subunits are required for wild type levels of germination.....	81

Figure 3.2.	Complementation of the A-type nGR subunits of <i>B. anthracis</i>	83
Figure 3.3.	The B- and C-type nGR subunits of <i>B. anthracis</i> can not be complemented <i>in trans</i>	88
Figure 3.4.	Overexpression of the <i>gerL</i> genes does not cause a dominant negative effect.	91
Figure 3.5.	The A-type nGR subunits of <i>B. anthracis</i> demonstrate limited functional redundancy..	93
Figure 3.6.	Global alignment of <i>B. anthracis</i> GerK _A , GerS _A and GerL _A amino acid sequences	100
Figure 4.1.	Expression of the <i>B. anthracis</i> GerH proteins with small epitope tags.....	107

ABSTRACT

Bacterial spores are dormant, highly resistant cell types formed in response to nutrient deprivation and environmental stressors. When conditions are suitable for growth, spores can quickly convert back to actively replicating, vegetative cells through the process of germination. Germination is a required step in the infectious cycle of many spore-forming pathogens, including *Bacillus anthracis*, the etiologic agent of anthrax. Nutrient germinant receptors (nGRs) are packaged within the spore and initiate germination in response to specific nutrient molecules. The nGRs are comprised of three subunits: the A-, B-, and C-type proteins. Currently, the exact mechanism of action of these proteins is unknown. Therefore, the goal of this work was to improve our understanding of the structure of the nGR complex, and the role of the individual nGR proteins in triggering germination to provide insight into the overall mechanisms of germination initiation. Here, the first successful overexpression of the A- and B-type nGR proteins is described. Using this system, the membrane topology of the GerH_A, GerH_B and GerH_C proteins was determined and the data were used to create a model of the nGR protein topology in the inner membrane of the spore. Importantly, these data represent the first structural information of the A- and B-type nGR subunits and have provided insight into their roles in triggering

germination. In addition, it is demonstrated that the A-type subunits of the *B. anthracis* nGRs share some functional redundancy, influence ligand specificity, and may require co-expression with the B- and C-type proteins for proper function. This dissertation work has advanced our understanding of the structure and function of the individual nGR subunits, an important step toward determining the mechanism of action of these receptors. This information may lead to identification of novel decontamination methods for preventing infections by the spore-forming pathogens.